

Lint development and properties of fifteen fuzzless seed lines of Upland cotton (*Gossypium hirsutum* L.)

Rickie B. Turley · Kevin C. Vaughn ·
Jodi A. Scheffler

Received: 24 June 2006 / Accepted: 29 December 2006 / Published online: 25 January 2007
© Springer Science+Business Media B.V. 2007

Abstract Fifteen fuzzless seed lines in “obsolete” backgrounds of Upland cotton (*Gossypium hirsutum* L.) were obtained from the National Cotton Germplasm Collection and evaluated for fuzzless seed genotype, lint percent, and lint quality. Fourteen of these fifteen lines were found to be homozygous for the dominant fuzzless seed allele N_1 . Only one line was homozygous for the recessive fuzzless seed allele n_2 . The measured lint percent of each line was very stable through time, however, large variability existed between many of the N_1 lines ranging from 0.7 to 23.6% lint. The lint percent for the n_2 line was 24.4%. Scanning electron microscopy was used to differentiate patterns of lint initiation on 1 day post anthesis ovules. General patterns included: first,

lint initiation restricted to the chalazal end of the seed crest; second, lint initiation along the seed crest and laterally around the chalazal end of the ovule; and third, lint initiation covered all but the micropylar end of the ovule. Lint quality was evaluated for each line using the Advanced Fiber Information System (AFIS) which included measurements of length, maturity, and fineness. The means of each measurement varied among the fuzzless seed lines with significant differences between fiber length, short fiber content, immature fiber content, fineness, and maturity ratio. No correlation was found between lint percent and any of the lint quality measurements. These lines will provide a valuable resource for the study of fiber initiation and lint quality.

Mention of trade names or commercial products in this manuscript is solely for the purpose of providing specific information and does not imply recommendation or endorsement by the U.S. Department of Agriculture.

R. B. Turley (✉) · J. A. Scheffler
Crop Genetics and Production Research Unit,
USDA-ARS, P.O. Box 345, Stoneville,
MS, 38776, USA
e-mail: rturley@msa-stoneville.ars.usda.gov

K. C. Vaughn
Southern Weed Science Research Unit,
USDA-ARS, P.O. Box 350, Stoneville,
MS, 38776, USA

Keywords AFIS · Fiber · Fuzzless seed ·
Gossypium hirsutum · Initiation · Naked seed

Abbreviations

2.5SL	2.5% span length
5.0SL	5.0% span length
AFIS	Advanced fiber information system
DPA	Days post anthesis
F	Fuzzy seed
Fine	Mean fiber fineness (mTex)
GRIN	Germplasm resources information network
IFC	Immature fiber content

$L(n)$	Mean length of fiber calculated on basis of fiber number
$L(w)$	Mean length of fiber calculated on basis of fiber weight
N	Fuzzless seed
MR	Maturity ratio
SFN	Short fiber content less than 12.7 mm calculated on a fiber number basis
SFW	Short fiber content less than 12.7 mm calculated on a fiber weight basis
UQL(w)	Upper quartile length

Introduction

Cotton is a highly valued natural fiber which accounts for approximately \$120 billion per year business revenue in the United States of America (National Cotton Council 2004). Therefore, improvements to both the yield and lint quality of cotton are research priorities. On a normal cottonseed two types of fiber are produced, lint and fuzz. Lint fibers initiate growth between anthesis and five days post anthesis (DPA) and can elongate to 2.5–3.5 cm (Seagull and Giavalis 2004; Stewart 1975). These are the economically important fibers. Fuzz fibers initiate growth by 10 DPA and elongate to approximately 0.5 cm (Seagull & Giavalis, 2004; Stewart, 1975). Two naturally occurring mutations are found in cotton which permit the development of lint fiber but prevent development of fuzz fiber. These are referred to as the fuzzless seed (also known as “naked seed”) alleles and designated as genotypes N_{1-} or n_2n_2 (Percy and Kohel 1999; Turley and Kloth 2002). The fuzzless seed lines provide unique tools to evaluate the effects of the fuzzless seed genotypes on fiber initiation, lint production and quality.

It was reported previously that the dominant fuzzless seed allele N_1 was genetically linked to lower lint percent (Thadani 1923; Kearney and Harrison 1927). A review of the Germplasm Resources Information Network (GRIN), United States Department of Agriculture, Agricultural Research Services database for the fifteen fuzzless seed lines reported in this study revealed that large differences existed between their lint percents, with values ranging from 1.9% to 25.8%.

The GRIN database, however, provided no information on the genotype, where these plants were grown, or how many replications were used to determine the lint percent of each line. The large deviation in lint percent indicates that other genetic factors which repress or enhance the lint percent may exist in these lines.

Genotypes of two of the fifteen lines in this study have been previously reported in the literature, seed accessions (SA's) 143 which is homozygous for n_2 and SA 243 which is homozygous for N_1 (Turley and Kloth 2002). Verifying the genotype for the other 13 lines becomes a priority in order to associate a genotype with measurements of lint development or quality. These lines were re-evaluated for lint percent over three summers and scanning electron microscopy was used to evaluate lint initiation patterns on each line. Ten lint quality measurements using the Advanced Fiber Information System (AFIS) were assessed for each line including length, short fiber content, immature fiber content, fineness, and maturity ratio. Correlations between lint properties and lint percent were also determined.

Materials and methods

Plant material and genetics

Fifteen inbred lines reported to express the fuzzless seed phenotype were obtained from the National Cotton Germplasm Collection in the Mississippi Obsolete Variety Collection subset managed by the US National Collection of *Gossypium* Germplasm (Percival 1987). In 2002, these fifteen SA's were grown in the field to verify the fuzzless seed phenotype and increase the seed for each line. Two of the SA's, 143 and 243, have been previously characterized and used in the development and test crossing of the fiberless MD 17 line (Turley 2002; Turley and Kloth 2002). The other thirteen SA's (GRIN numbers reported in Table 1) were 27, 48, 50, 51, 52, 53, 66, 68, 128, 176, 177, 421 and 505. The conventional cultivar DP 5690 (Delta and Pine Land, Scott, MS, USA) also was included in this study to compare with the fuzzless seed lines.

Table 1 Summary of the seed accessions (SA), GRIN numbers, reported lint percents in the GRIN database, and measured lint percents at Stoneville, MS (values in column followed by the same letter are not significantly different at the 0.05 level as determined by the LSD). All F₁ and F₂populations were derived from crosses of DP 5690 with the individual “fuzzless seed” line listed in the specified row. Best fit models were determined using χ^2 . Genotypes and Phenotypes are also listed with the Table separated into three groupings according to genotype

SA	GRIN	Reported Lint Percent	Measured [‡] Lint Percent	Number of Plants in F ₁ Populations		Number of Plants in F ₂ Populations		χ^2 1F:3N	Genotype	Seed Phenotype
				F ^A	N ^A	F	N			
27	PI 528444	16.2	17.1 D	0	53	118	358	0.0112	$N_1N_1N_2N_2$	Fuzzless
48	PI 528461	NR ^A	0.7 I	0	46	93	303	0.4848	$N_1N_1N_2N_2$	Fuzzless
50	PI 528463	23.6	15.9 D	0	25	91	277	0.0145	$N_1N_1N_2N_2$	Fuzzless
51	PI 528464	1.9	2.3 HI	0	41	86	260	0.0039	$N_1N_1N_2N_2$	Fuzzless
52	PI 528465	25.8	23.6 B	0	47	96	295	0.0418	$N_1N_1N_2N_2$	Fuzzless
53	PI 528466	19.3	19.4 C	0	47	110	323	0.0377	$N_1N_1N_2N_2$	Fuzzless
66	PI 528477	3.9	3.4 GH	0	53	115	337	0.0472	$N_1N_1N_2N_2$	Fuzzless
68	PI 528479	10.2	10.9 E	0	49	98	299	0.0210	$N_1N_1N_2N_2$	Fuzzless
128	PI 528531	8.9	5.9 F	0	57	96	309	0.3630	$N_1N_1N_2N_2$	Fuzzless
176	PI 528569	NR	3.3 GH	0	49	104	333	0.3364	$N_1N_1N_2N_2$	Fuzzless
177	PI 528570	3.8	3.2 H	0	16	86	285	0.6550	$N_1N_1N_2N_2$	Fuzzless
243	PI 528610	11.4	11.6 E	–	–	–	–	–	$N_1N_1N_2N_2$	Fuzzless
421	PI 528737	6.4	5.3 FG	0	56	79	254	0.2893	$N_1N_1N_2N_2$	Fuzzless
505	PI 528808	14.2	10.7 E	0	39	86	240	0.3313	$N_1N_1N_2N_2$	Fuzzless
143	PI 528543	25.6	24.4 B	–	–	–	–	–	$n_1n_1n_2n_2$	Fuzzless
5690	NR	NR	37.7 A	–	–	–	–	–	$n_1n_1N_2N_2$	Fuzzy

^A Abbreviations include: F for fuzz covered seed, N for fuzzless seed, and NR for not reported[‡] The LSD for the measured lint percent was 1.977

This study occurred over three years, 2002 to 2004, at Stoneville, MS, USA. All studies were organized in randomized complete blocks with three replications. Field plots were single row, five meters long and spaced 1.02 m apart. Where applicable, plots were over-seeded and after the plants reached the first true leaf stage, seedlings were thinned to 6.5 plants m⁻². Weeds and insects were managed using standard agronomic practices for the Mississippi delta.

The thirteen uncharacterized fuzzless seed lines (pollen parent) were crossed with DP 5690 in 2002 and the F₁ generations were self pollinated in a greenhouse (Jan–Feb 2003). The F₂ populations were evaluated for the fuzzless seed phenotype in 2003 and 2004. The fuzzy and fuzzless phenotypes were scored as described by Turley and Kloth (2002) by examining seeds from open capsules at the first branch node between main stem nodes seven through eleven. Chi square values were calculated to determine the best fit for all genetic models tested. The

models tested for the segregation ratios of fuzzy seed (F) to fuzzless seed (N) were 1F:3N for the dominant N_1 allele and either 3F:1N or 15F:1N for the recessive n_2 allele (Turley and Kloth 2002).

Determination of lint percent and lint quality

Lint percent was calculated by dividing the mass of lint ginned by the mass of seed cotton (total weight of lint and seed) and expressed as a percentage of the mass of seed cotton. Lint percents were calculated for the fuzzless seed lines and DP 5690 by harvesting all seed cotton from a single plot. Lint from the fuzzless seed lines was removed manually due to the low amount of lint produced on a few lines. Previously reported lint percents for these lines, found at http://www.ars-grin.gov/cgi-bin/npgs/html/tax_site_acc.pl?COT%20Gossypium%20hirsutum, were compared with the measured lint percents. Low lint yields, especially for SA 48, limited options for lint analysis to AFIS (Calhoun et al.

1997). All references to specific fiber measurements obtained from AFIS refer explicitly to lint fiber.

Lint analyses were performed on an USTER AFIS™ (ZellwegerUster, Uster, Switzerland). Lint measurements included: mean length (mm) of the fiber calculated on basis of fiber number, $L(n)$; mean length (mm) of fiber calculated on basis of fiber weight, $L(w)$; upper quartile length calculated on basis of fiber weight, $UQL(w)$; 5.0% span length, 5.0SL (fiber length exceeded by only 5.0% of the fibers); 2.5% span length, 2.5SL (fiber length exceeded by only 2.5% of the fibers); percentage of short fiber content less than 12.7 mm calculated on a fiber number basis, SFN; percentage of short fiber content less than 12.7 mm calculated on a fiber weight basis, SFW; mean fiber fineness or mTex, Fine; immature fiber content, IFC; and maturity ratio, MR. MR is the ratio of fibers with a 0.50 or more circularity divided by the amount of fiber with a 0.25 or less circularity (Williams and Yankey 1996; Calhoun et al. 1997).

Lines were compared with respect to lint percent and lint quality using Fisher's protected LSD with $P < 0.05$ reported. Average LSD's are reported for all lint quality measurements due to the lack of one replication for SA 143 in 2002 and for SA 177 in 2003. Pearson's correlations were calculated to compare the means of lint percent and lint quality measurements. All statistical analyses were performed using SAS (SAS Institute, Cary, NC, USA).

Scanning electron microscopy (SEM)

One DPA ovules from the sixteen lines were fixed in 5 ml of 6% glutaraldehyde in 50 mM Pipes (pH 7.4) overnight at 4°C. Samples were rinsed twice in 10 mM cacodylate (pH 7.2) for 15 min. An additional two rinses were performed with deionized water. The samples were then dehydrated in an ethanol series: 25% for 15 min, 50% for 15 min, 75% for 15 min. The 75% solution was then decanted, 100% ethanol was added, and the samples dehydrated overnight at 4°C. The ethanol was removed and the ovules were critical point dried substituting carbon dioxide to remove all traces of water. Eight dried ovules from each line were mounted on aluminum stubs and sputter

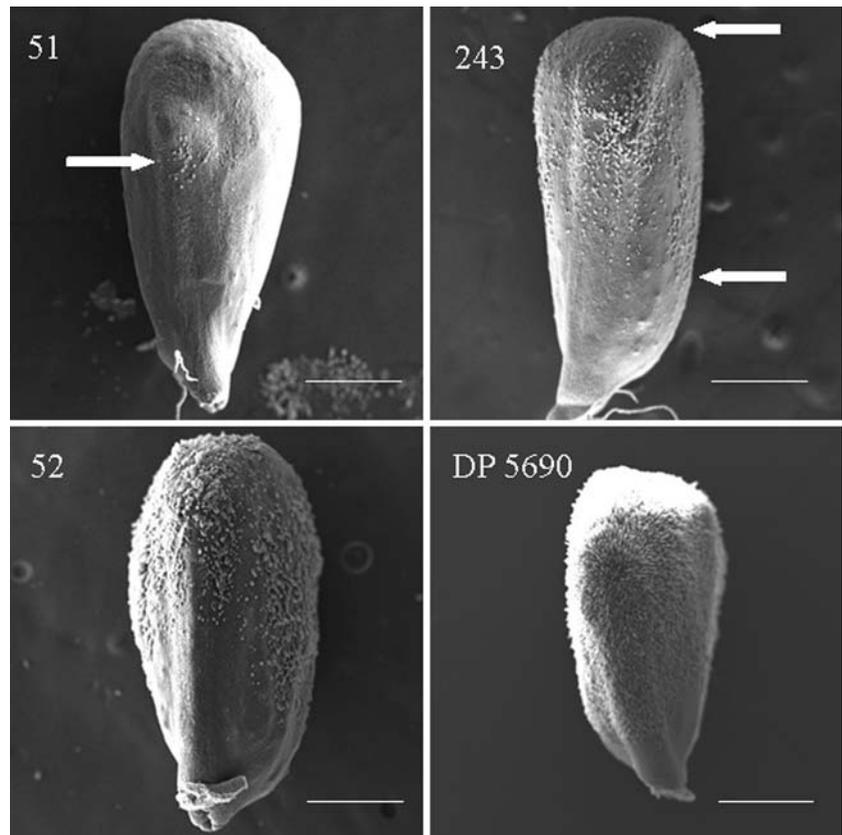
coated (Hummer X Coater, Anatech LTD., Denver, NC, USA) with 18 nm of 100% gold-palladium. Samples were viewed using a Jeol 840 scanning electron microscope.

Results and discussion

Reported and measured lint percents are summarized in Table 1. Table 1 is organized sequentially by SA numbers of the N_1N_1 lines, then n_2n_2 and 5690. As expected, most of the measured lint percents were very similar to the reported lint percents, however, for three SA's, 50, 128, and 505, the measured lint percents were at least 25% lower than the reported lint percents. Two other SA's, 48 and 176, did not have reported lint percents in the GRIN database, therefore, the measured lint percents are the first reports in the literature (Table 1).

Because of the wide variability of measured lint percents (from 0.7 to 23.6% lint), scanning electron microscopy was used to confirm whether lint patterns on the fuzzless seed lines were exclusively due to fiber initiation, or to fiber initiation followed by localized apoptosis. After an evaluation of all SA lines, micrographs of four representative 1 DPA ovules were selected to demonstrate general patterns of lint initiation (Fig. 1). Seed accessions 48 and 51 had the lowest lint percents and similar patterns of lint initiated localized exclusively at the chalazal end of the seed crest (see arrow in SA 51, Fig. 1). Mature seeds of SA's 48 and 51 had patterns of lint distribution equivalent to the lint initiation pattern on the 1 DPA ovules. This was manifested as a chalazal tuft on the mature seed (data not shown). As the lint percents increased, the area of lint initiation also increased laterally around the chalazal end of the ovule (represented by SA 243, Fig. 1). For the SA's 176, 66, 421, 128 and 68 with lower lint percents than SA 243, the patterns were less dense and generally covered about one third of the ovule on the chalazal end. SA 243 at 11.6% lint has lint which covers approximately two thirds of the ovule (see arrows in SA 243, Fig. 1). SA 52 produces the most lint of the N_1 lines at 23.6% lint with about two-thirds the ovule covered (Fig. 1). For all SA's, lint initiation patterns on

Fig. 1 Scanning electron micrographs of four representative ovules (seed accessions 51, 243, 52 and the cultivar DP 5690). SA 51, arrow indicates initiating lint. SA 243, two arrows delineate the area of lint initiation. The bar at the bottom right hand corner of each micrograph equals 0.5 mm



one DPA ovules persisted to mature cottonseeds (data not shown). No quantitative measurements were attempted to verify whether there was a higher lint density on the mature seeds with higher lint percents. The DP 5690 cultivar had lint growth covering most of the ovule (Fig. 1).

The 1 DPA time point was selected because previous work on DP 5690 in our laboratory indicated that by this time point the ovule was covered with lint (Fig. 1). Lint in the fuzzless seed lines in most cases appear shorter and less developed. (51, Fig. 1) than in the DP 5690 line (data not shown). Care was taken to harvest all the cotton capsules from developmentally similar positions on the plant. The relative slower fiber growth in the N_1 lines was recently confirmed in a recent report by Lee et al. (2006) using a near isogenic N_1 in a TM-1 background. Seagull and Giavalis (2004) found that lint initiation can occur as late as 5 DPA, well after the 1 DPA used in this report. Additional lint initiation in the fuzzless seed lines probably occurred, but was likely

restricted to the generalized patterns discussed above with only modest gains in fiber covering the surface on the mature seed. This assumption was made based on the fact that lint initiation patterns on 1 DPA ovules were equivalent to lint development patterns on mature seeds. SA's 48 and 51 only produced lint at the chalazal end of the seed crest. Moore (1941) reported that lint initiation at the chalazal end of the seed crest was five times greater than any other area on the seed (measured as mature lint). No evidence was ever found indicating lint initiated growth then rapidly progressed through apoptosis.

We have previously reported that SA 143 was homozygous for the recessive n_2 allele and SA 243 was homozygous for the dominant N_1 allele (Turley and Kloth 2002). To determine the genotype of the remaining thirteen SA's, crosses were made between DP 5690 X "SA" (each of the thirteen fuzzless seed lines). Each of the thirteen previously non-genotyped fuzzless seed lines listed in Table 1 was determined to be homozygous for the

N_1 allele. This conclusion was reached because the F_1 populations were 100% fuzzless seed and the F_2 populations segregated in a 1F:3N ratio. Both results indicated the presence of the dominant N_1 allele. The probability of n_2 allele involvement in the expression of the fuzzless phenotype for any of these SA's was completely eliminated when Chi square analysis was used to evaluate the recessive segregation ratios of 3F:1N and 15F:1N in the F_2 population.

The fuzzless seed lines provide a unique opportunity to study lint initiation on cotton ovules. Fourteen of the fifteen fuzzless seed lines evaluated in this study were homozygous for the N_1 fuzzless seed allele which was previously reported to be linked to lower lint percent in cotton (Thadani 1923; Kearney and Harrison 1927). The large variation in lint percent from 0.7 to 23.6% lint suggests one of two scenarios. The first scenario would be that the expression of other alleles interact with the N_1 allele either positively, or negatively, to modify lint percent. The second scenario would indicate that multiple N_1 alleles exist in cotton.

The first scenario has already been shown to occur in cotton, in that fiberless lines MD 17 and XZ142w need additional modifying loci to completely inhibit lint development (Du et al. 2001; Turley 2002; Turley and Kloth 2002; Zhang and Pan 1991). Modifying loci which increase lint percent have not been reported, however, preliminary evidence suggests that loci which can increase the lint percents in N_1 lines may actually exist. The N_1 allele from SA 243 was recently transferred into a near isogenic DP 5690 background using the backcrossing method through five generations (Turley, germplasm release pending). This near isogenic line was homozygous for N_1 and had a lint percent of 11.5%, equivalent to its original N_1 donor parent SA 243 (Table 1). Assuming that SA 243 and DP 5690 did not possess a similar/identical inhibitor of the N_1 allele, then the significantly higher lint percents for SA's 50 (15.9% lint), 27 (17.1% lint), 53 (19.4% lint), and 52 (23.6% lint) indicate that modifier genes may exist in these genetic backgrounds which increase the amount of lint produced.

The second scenario for the higher lint percents could be explained by multiple N_1 alleles that pos-

sibly exist in cotton. No records exist as to whether these fuzzless SA's were found as spontaneous mutations, or whether they were derived by crossing the N_1 allele into these "obsolete" backgrounds. Different mutations in the N_1 allele could possibly produce lines with higher lint percents than the N_1 allele from SA 243 when expressed in the same near isogenic line. Further work is presently underway to test these two scenarios by using allelic analyses and near-isogenic lines.

Lint quality measurements for each SA and DP 5690 are reported as a mean of three years of analysis (Table 2). Table 2 is organized differently than Table 1, in that Table 2 is ranked based on lint percent with the line producing the lowest lint percent at the top (SA 48, 0.7% lint) to the line producing the highest lint percent at the bottom (DP 5690, 37.7% lint). The N_1 lines are grouped for facilitated comparisons. SA 48, an almost fiberless line, had the shortest fiber $L(n)$ and $L(w)$, the highest short fiber and immature fiber content (SFN, SFW and IFC), the second finest fiber (Fine), and the lowest maturity ratio (MR). Three other SA's, 27, 53 and 52 each deserve a note with some unique lint qualities. SA 27 with 17.1% lint has the longest fiber measurements $L(n)$, $L(w)$, UQL(w), 5.0SL and 2.5SL, with the second finest fiber (Fine). SA 53 with 19.4% lint has the shortest fiber at the UQL(w), 5.0SL and 2.5SL levels, the lowest short fiber and immature fiber content (SFN, SFW, and IFC), and the highest Fine and MR values. SA 52 has the highest lint percent of the N_1 lines at 23.6% lint but has only average fiber quality measurements (Table 2). DP 5690, which was the only seed cotton which was mechanically ginned in the group, provided a comparison of lint measurements with a conventional variety.

The lint quality measurements in Table 2 were also variable with large statistical differences between measurements. No correlations were found between lint percent and any of the other lint quality measurement. Numerous correlations were identified between other measurements of lint quality as reported in Table 3. Lint length measurements $L(n)$, $L(w)$, UQL(w), 5.0SL and 2.5SL were highly correlated as expected (Table 3). Fiber fineness (Fine) was negatively correlated with four length [$L(w)$, UQL(w), 5.0SL

Table 2 Mean lint measurements for fuzzless seed accessions (SA) and the commercial cotton cultivar DP 5690. Values in a column followed by the same letter are not significantly different at the 0.05 level as determined by the LSD. LSD (Average LSD) is also reported for each fiber measurement at the $P < 0.05$. Table separated into three categories according to genotype

SA	$L(n)^a$	$L(w)$	UQL(w)	5.0SL	2.5SL	SFN	SFW	Fine	IFC	MR
48	17.8 F	22.2 G	26.3 GH	29.8 GH	31.7 GHI	27.7 A	8.6 A	191.6 B	7.5 A	0.86 G
51	21.1 DE	24.0 EF	27.4 EFG	31.1 EFG	33.1 CDEFGH	13.5 BCDE	3.9 BCDEF	208.9 CD	4.5 BC	0.95 DEF
177	23.2 BCD	25.5 BCDE	28.7 CDEF	32.4 CDEF	34.5 BCDE	8.9 CDEFG	2.6 CDEFG	203.9 BCD	3.9 BCDE	0.98 BCDE
176	20.6 E	23.2 FG	26.4 G	29.8 GH	31.7 GHI	13.5 BCDE	4.2 BCDE	218.0 ED	4.1 BCD	0.96 CDEF
66	21.7 CDE	25.7 BCD	30.0 BC	33.9 BC	36.0 B	18.9 B	5.3 B	189.7 B	5.5 B	0.92 F
421	23.7 AB	26.2 BC	29.5 CD	33.3 CD	35.1 BC	9.1 EFG	2.2 EFG	211.2 CDE	3.4 CDE	0.99 BCD
128	21.1 DE	24.6 DEF	28.1 DEF	31.9 DEF	34.0 BCDEF	16.3 BC	4.4 BCD	195.8 BC	5.0 B	0.93 F
505	22.1 BCDE	23.9 EF	27.0 FG	31.1 EFG	32.5 EFGH	7.6 EFG	2.6 DEFG	226.8 E	2.6 DE	1.00 BC
68	21.3 CDE	23.2 FG	26.2 GH	29.7 GH	31.3 HI	9.4 DEFG	3.2 CDEFG	244.2 F	3.0 CDE	0.98 BCDE
243	22.0 BCDE	25.4 BCDE	29.0 CDE	32.8 CDE	34.8 BCD	16.1 BCD	4.7 BC	198.1 BC	5.0 B	0.92 F
50	23.2 BC	25.1 CDE	28.2 DEF	31.8 DEF	33.5 CDEFG	7.3 EFG	2.2 EFG	221.2 DE	3.1 CDE	1.00 BC
27	25.6 A	29.2 A	33.7 A	38.1 A	40.5 A	12.2 CDEF	3.0 CDEFG	188.6 B	4.4 BC	0.95 CDEF
53	21.3 CDE	22.4 G	24.8 H	28.1 H	29.7 I	3.9 G	1.5 G	245.1 F	1.1 F	1.06 A
52	22.4 BCDE	24.6 DEF	27.8 EFG	31.2 EFG	33.0 DEFGH	10.1 CDEFG	3.2 CDEFG	208.6 CD	4.5 BC	0.94 EF
143	22.8 BCDE	24.2 DEF	26.9 FG	30.1 FGH	31.8 Fghi	4.9 FG	1.6 FG	229.2 EF	2.2 EF	1.03 AB
5690	22.8 BCD	26.7 B	31.5 B	35.8 B	38.5 A	15.9 BCD	4.8 BC	171.3 A	4.1 BCD	0.97 CDE
LSD	2.075	1.523	1.689	1.958	2.128	6.888	2.154	17.35	1.538	0.047

^a Mean length of fiber calculated on basis of fiber number, $L(n)$; mean length of fiber calculated on basis of fiber weight, $L(w)$; upper quartile length, UQL(w); 5.0% span length, 5.0SL; 2.5% span length, 2.5SL; short fiber content less than 12.7 mm calculated on a fiber number basis, SFN; short fiber content less than 12.7 mm calculated on a fiber weight basis, SFW; mean fiber fineness, mTex, Fine; immature fiber content, IFC; and maturity ratio, MR

Table 3 Summary of Pearson's correlation coefficients between fiber measurements for all sixteen lines over a three year period

	L(n) ^a	L(w)	UQL(w)	5.0SL	2.5SL	SFN	SFW	Fine	IFC	MR
L(n)	–									
L(w)	0.836**	–								
UQL(w)	0.689**	0.972**	–							
5.0SL	0.676**	0.964**	0.997**	–						
2.5SL	0.637**	0.950**	0.995**	0.997**	–					
SFN	–0.551*	–0.009	0.209	0.218	0.263	–				
SFW	–0.659**	–0.150	0.074	0.085	0.131	0.856**	–			
Fine	–0.093	–0.595*	–0.742**	–0.748**	–0.783**	–0.713**	–0.621*	–		
IFC	–0.407	0.111	0.292	0.292	0.328	0.944**	0.901**	–0.750**	–	
MR	0.411	–0.080	–0.250	–0.250	–0.281	–0.910**	–0.869**	0.680**	–0.984**	–

*, ** significant at the 0.05 and the 0.01 levels, respectively

^a Measurements tested included: mean length of fiber calculated on basis of fiber number, $L(n)$; mean length of fiber calculated on basis of fiber weight, $L(w)$; upper quartile length, UQL(w); 5.0% span length, 5.0SL; 2.5% span length, 2.5SL; short fiber content less than 12.7 mm calculated on a fiber number basis, SFN; short fiber content less than 12.7 mm calculated on a fiber weight basis, SFW; mean fiber fineness, mTex, Fine; immature fiber content, IFC; and maturity ratio, MR

and 2.5SL], two short fiber (SFN and SFW) and immature fiber content (IFC) measurements (Table 3). Fiber fineness (Fine) was positively correlated with fiber maturity ratio (MR). At this point, it is impossible to propose any association between the N_1 allele and determinations of lint quality, however, it was not found that the homozygous expression of N_1 had adverse effects on fiber length (Table 2) as was recently reported by Lee et al. (2006).

These N_1 lines offer a unique tool to evaluate lint development without the complication of fuzz fiber production. Collectively, the data obtained from these lines delineate the large diversity of the fuzzless seed lines that exist in the National Cotton Germplasm Collection. Because fourteen of the fifteen fuzzless seed lines were homozygous for the N_1 allele with lint percents ranging from 0.7% to 23.6%, we now have an experimental base in which to identify and evaluate either negative or positive modifiers of lint percent in the N_1 lines. The possibility of modifier genes that increase lint percent could be extremely important to the cotton industry because yield potential of cotton in recent years has reached a plateau (Meredith 1995). Testing and proving the existence of these modifiers will take time; incorporating possible modifiers from these lines (SA's 50, 27, 53, and 52) into a breeding program to verify possible increases in lint percents has already begun.

These fuzzless seed lines are also a valuable resource in the study of trichome (lint) initiation and development. The lower lint percents make these lines ideal for studies in lint density and spacing on the cottonseed. Generalized patterns of lint initiation were identified and reported above. Development of near isogenic lines with similar initiation patterns and lint percents to the above lines would greatly benefit researchers in the cotton community. The authors are aware of numerous funded projects presently underway which utilize a fuzzless seed line as a primary tool in the investigation of lint development. As the data from this study indicate, not all fuzzless seed lines are the same, and therefore, care must be taken in the interpretation of the results.

Acknowledgments We would like to thank William Nokes, Rex Manning, Cedric Brown, Tony Miller and Dr. William Pettigrew for assistance in field preparation and irrigation; Dr. Jeffrey Ray for his program to determine chi square values; Shirley Howard, Flossie Warfield and Elnora Trotter for sample preparation for AFIS analysis; Brian Maxwell for sample preparation for SEM analysis; Debbie Boykin for her statistical evaluation; and to Drs. Sally Stetina and Gayle Davidonis for critically reviewing this manuscript

References

- Calhoun DS, Barger JD, Anthony WS (1997) An introduction to AFIS for cotton breeders. In: Dugger P, Richter DA (eds) Proc beltwide cotton conf, National Cotton Council, New Orleans, LA, USA, pp 418–424

- Du XM, Pan JJ, Wang RH, Zhang TZ, Shi TZ (2001) Genetic analysis of presence and absence of lint and fuzz in cotton. *Plant Breed* 120:519–522
- Kearney TH, Harrison RJ (1927) Inheritance of smooth seed in cotton. *J Agric Res* 35:193–217
- Lee JJ, Hassan OSS, Gao W, Wei NE, Kohel RJ, Chen XY, Payton P, Sze SH, Stelly DM, Chen ZJ (2006) Developmental and gene expression analyses of a cotton naked seed mutant. *Planta* 223:418–432
- Meredith WR (1995) Strengths and limitations of conventional and transgenic breeding. In: Richter DA, Armour J (eds) *Proc Beltwide Cotton Conf*, National Cotton Council, San Antonio, TX, USA, pp 166–168
- Moore JH (1941) The distribution and relation of fiber population, length, breaking load, weight, diameter, and percentage of thin walled fibers on the cottonseed in five varieties of American upland cotton. *J Agric Res* 62:255–303
- National Cotton Council (2004) <http://www.cotton.org/econ/world/index.cfm>
- Percival AE (1987) The national collection of *Gossypium* germplasm. *Southern Cooperative Series Bul* 321
- Percy RG, Kohel RJ (1999) Qualitative genetics. In: Smith CW, Cothren JT (eds) *Cotton: origin, history, technology, and production*, John Wiley and Sons, New York, NY, pp 319–360
- Seagull RW, Giavalis S (2004) Pre- and Post- anthesis application of exogenous hormones alters fiber production in *Gossypium hirsutum* L. cultivar Maxxa GTO. *J Cotton Sci* 8:105–111
- Stewart JM (1975) Fiber initiation on the cotton ovule (*Gossypium hirsutum*). *Amer J Bot* 62:723–730
- Thadani KI (1923) Linkage relationships in the cotton plant. *Agr J India* 18:37–42
- Turley RB (2002) Registration of MD 17 fiberless upland cotton as a genetic stock. *Crop Sci* 42:994–995
- Turley RB, Kloth RH (2002) Identification of a third fuzzless seed locus in upland cotton (*Gossypium hirsutum* L.). *J Hered* 93:359–364
- Williams GF, Yankey JM (1996) New developments in single fiber fineness & maturity measurements. In: Dugger P, Richter DA (eds) *Proc Beltwide Cotton Conf*, National Cotton Council, Nashville, TN, USA, pp 1284–1289
- Zhang TZ, Pan JJ (1991) Genetic analysis of a fuzzless-lintless mutant in *Gossypium hirsutum* L. *Jiangsu J Agric Sci* 7:13–16